A REPORT ON THE USE OF A PECTORAL MUSCLE BIOPSY IN THE FIELD FOR ORGANOCHLORINE RESIDUE ANALYSIS*

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Introduction

In the course of a survey of organochlorine residues in East African raptors in 1971-72, we found that a large proportion of trapped birds had insufficient fat reserves to allow a subcutaneous fat biopsy (Enderson and Berger 1968). As we were unwilling to collect raptors to take tissue samples, we decided to adopt the method of Seidensticker (1970) for taking biopsies of the pectoral muscle, as this technique is no more complex or time-consuming than the fat biopsy. He reported no significant impairment in captive Red-tailed Hawks (*Buteo jamaicensis*) and Golden Eagles (*Aquila chrysaetos*), and, after testing it on captive birds in Kenya, we used the technique on 10 birds that were subsequently released.

Methods

To obtain the sample, the bird is hooded and held by an assistant while it is injected with anesthetic; we at first used metomidate (Hypnodil, Janssen Pharmaceutica) but found that this agent kept the bird sedated far longer than necessary to accomplish the biopsy, and excessive salivation necessitated constant swabbing of the pharynx to prevent choking and gagging on saliva. We then switched to CT 1341 (Saffan, Glaxo Ltd.) (Cooper and Frank 1973) which proved ideal, as it induces surgical anesthesia immediately upon intravenous injection, causes no salivation or other complications, and the bird is fully recovered (responds normally to stimuli, able to stand, perch, and fly) within 30 or 40 minutes of injection.

The anesthetized bird is laid on its back, the keel region wetted with ethanol, and the feathers parted to expose a patch of skin about 2 cm wide by 3 cm

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long lateral to the most ventral point of the sternum; the size of the bared patch depends upon the size of the bird, and a few feathers can be plucked if necessary. After the skin has been cleaned with an alcohol swab, an incision 2.5 cm long is made 8-10 mm lateral to the ridge of the sternum, and the skin spread apart to expose the pectoralis muscle and overlying fat (if any). An incision is then made in the muscle parallel to the keel, 2 cm long and 3-4 mm deep; this incision will widen as a result of muscle tonus. A second incision is then made in the muscle, parallel and 4 mm dorsolateral to the first and angled toward it, to free a prism-shaped strip of muscle which can be lifted out with forceps. These measurements refer to a bird the size of a Golden Eagle and can be adjusted to a larger or smaller bird.

If performed on the suggested site on the breast, there is little or no hemorrhage; the few drops of blood that may appear can be removed with an alcohol sponge and a few seconds of light pressure with the sponge will stop the bleeding. The entire wound is then sprinkled with tetracycline powder, and the skin sutured by two or three mattress sutures, using size 00 gut. The sutured wound is then smeared with furacin ointment or sprayed with calloidin wound dressing and the parted feathers laid over the bare skin. With a little experience, the entire procedure can be carried out in well under ten minutes.

Results

Unfortunately, we did not have an accurate balance available and were unable to weigh the biopsy samples. Seidensticker (1970) reported average weights of .03 g for Red-tailed Hawks and 0.5 g for Golden Eagles.

Sixteen birds of nine genera were biopsied; of these, 10 were released at the site of capture within three days of surgery, and six were kept in captivity for varying lengths of time. In five cases we were able to observe biopsied birds for several days or weeks after release: in only one instance was there any overt abnormal behavior or sign of ill-health; this was a White-backed Vulture (*Gyps africanus*) from which had been removed a biopsy of approximately .8 g. It had been anesthetized with metomidate and was released the next day, 22 hours after surgery. Vultures had proven exceptionally sensitive to metomidate (Houston and Cooper 1973) and this one appeared rather groggy and unstable, though it flew off about 0.5 km and perched awkwardly in an accia tree. It stayed in the vicinity for the next .24 hours, but had flown off 48 hours after release. As vultures have taken as long as 72 hours to recover from metomidate, we felt that this one's reluctance to fly was due to the effects of the anesthetic rather than to pain from the biopsy wound.

In no other case did a released bird show any reluctance or inability to fly normally; of the six birds kept in captivity after surgery, four had previously been trained to fly at least five meters to the fist for food and none showed any hesitation to do so or to bate on the day following surgery. With the exception of the vulture, all birds observed after surgery, whether captive or released and observed subsequently, behaved normally and appeared healthy. Two immature African Fish Eagles (*Haliaeetus vocifer*) were seen near the point of release two

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and six weeks after surgery, and one Augur Buzzard (*Buteo rufofuscus*) was in attendance on its home territory for at least three months after release.

Discussion

The biopsy procedure as outlined above departs from the one described by Seidensticker only in the recommendation that the wound be sutured. He was using captive birds that were kept on perches and (presumably) fed dead food and he reported better wound healing and less infection if the incision in the skin was not sutured. On the contrary, we found that the wound healed more neatly if sutured, and felt that a hawk or eagle in the wild, exposed daily to the rough and tumble of capturing prey, stood a greater chance of getting foreign matter in an unsutured wound and, subsequently, a greater chance of infection. The single occurrence of infection in any of the captive birds we biopsied was in an unsutured Tawny Eagle (Aquila rapax) that was often recumbent due to a leg injury, and was therefore more likely to contaminate the wound than a healthy bird perched upright. This was the single case of complications in any of the wounds we observed after biopsy, and we felt it was sufficient grounds to suture any bird that may be exposed to foreign matter at the site of injury. As this possibility can never be eliminated, we recommend that the wound always be sutured, leaving at most a 3 mm gap at the posterior end of the wound for drainage.

The position of the incision is important, because the skin at the suggested site is not heavily vascularized, and hemorrhage is minimal. In the one instance when we departed from this site and made the incision further posterio-lateral on a Lanner Falcon (*Falco biarmicus*), there was heavy cutaneous bleeding that could have resulted in the loss of the bird.

With the exception of the one Tawny Eagle, recovery in all birds kept captive after surgery was uneventful; wound healing was rapid, complete in ten days, and the sutures fell out or were absorbed in three or four weeks.

The one possible disadvantage to using the pectoral muscle biopsy in raptors such as falcons or accipiters which are entirely dependent on high speed precision flight in hunting is that slight damage to the pectoral muscle may temporarily impair their powers of flight sufficiently to decrease hunting success. While we had no evidence to support this possibility, we felt it advisable to keep and feed all biopsied birds for at least 24 hours, and at least 72 hours in the case of falcons and accipiters. Birds less dependent on powerful flight such as Tawny Eagles, chanting goshawks (*Melierax* spp.), Augur Buzzards, White-backed Vultures, and African Fish Eagles, were released the day after surgery. On one occasion, a Pale Chanting Goshawk (*Melierax canorus*) had to be released as soon as it had recovered from the anesthetic (CT 1341); it flew off immediately and perched nearby. It was observed hunting in its home range the next day.

Because there might be slight impairment of flying abilities that is of no great consequence in normal circumstances, but that may become significant in times of increased flying activity, as in the case of a bird that is migrating or feeding nestlings, it is probably not advisable to perform the pectoral muscle biopsy on birds in either of these circumstances until we have sufficient data to show that it has no effect on flying prowess or endurance even under conditions of stress. This could possibly be tested by performing surgery on a trained falcon and testing its ability to stoop at a lure or fly in a wind tunnel for prolonged sessions following biopsy.

Seidensticker (1970) reported organochlorine residue values obtained from biopsy samples to be more variable than whole muscle samples, and that the biopsies contained a higher percentage of extractable lipids. Biopsy values for DDT and metabolites were 17% lower than whole muscle samples in DDT-dosed Red-tailed Hawk fledglings, and he states that "... chlorinated hydrocarbon residues found in muscle samples which were taken via the biopsy should be viewed as estimates of the contents of the entire tissue rather than as absolute values." The present authors concur with this opinion, but propose that further experimental studies be undertaken with a larger sample size of dosed birds in the hope of establishing a relatively constant relationship between residue levels found in the biopsy samples and whole muscle.

Summary

The pectoral muscle biopsy technique reported by Seidensticker (1970) was used in captive and wild raptors of nine genera as part of a pesticide residue survey in Kenya. It proved equally simple to perform in the laboratory or under field conditions, and there was no discernible impairment of flying abilities in 15 of the 16 birds biopsied. In the one exception, the reluctance to fly was attributed to after-effects of the anesthetic rather than the biopsy, and the one instance of infection in a bird that was kept in captivity after surgery was apparently due to contamination of the unsutured wound. It is suggested that the wound always be sutured to prevent infection in released birds and the advisability of performing any muscle surgery on a bird that is migrating or feeding young is questioned. More data on the relationship between residue levels in biopsies and whole muscle are needed.

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